Mating type distribution and pathogenicity of Phytophthora infestans in Taiwan

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Abstract. Taiwanese isolates of Phytophthora infestans were examined for mating types and pathogenicity to tomato and potato. A total of 70 isolates, including 68 isolates from tomato and 2 isolates from potato, from six counties of Taiwan were all A1 mating type. Three tomato isolates of P. infestans caused only leaf blight of tomato, while two isolates obtained from potato were able to cause the disease both on potato and tomato, indicating that host-specific strains of P. infestans occur in Taiwan.

Keywords: Host specificity; Mating type; Phytophthora infestans; Potato; Taiwan; Tomato.

Introduction

The fungus Phytophthora infestans (Montagne) de Bary is a heterothallic oomycete with two compatibility mating types designated as A1 and A2 (Gallegly and Galindo, 1960). This species has been known to exist in Taiwan since the early 1900s, causing late blight of potato and tomato (Kawakami and Suzuki, 1908; Sawada, 1919). Three tomato isolates of the fungus belonging to the A1 mating type from Taiwan have been analyzed for genetic variation at two allozyme loci, glucose-6-phosphate isomerase and peptidase, and for nuclear DNA haplotype. The three isolates all had a genotype identical with the one previously found throughout the world, designated as US-1 (Koh et al., 1994). However, the mating type composition and pathogenicity to tomato and potato within the P. infestans population of Taiwan have not previously been documented. Results of an investigation into these aspects are reported in this paper.

Materials and Methods

Isolation of Phytophthora infestans

Samples of blighted tomato and potato leaves were collected from tomato and potato fields in 1993–1996. The blighted leaves were kept in a plastic bag for 2–4 days at 20°C to promote sporangia formation. The sporangia were picked off the blighted leaves using a sterile needle and spread on rye B agar (Frinking et al., 1987) or green pea agar (Shattock et al., 1990) amended with 100 ppm ampicillin, 50 ppm mycostatin, and 10 ppm pentachloronitrobenzene after autoclaving. After incubation at 20°C for 5–10 days, mycelia of P. infestans germinating from sporangia were transferred to rye B agar or green pea agar.

Characteristics of Phytophthora infestans

Sporangia for measurement were obtained from cultures of P. infestans grown on rye B agar for 2–3 weeks in darkness. P. infestans isolates were grown on rye B agar for 5–10 days. Agar disks (5 mm in diameter) cut from the periphery of the colonies with a sterile cork borer were used to inoculate plates. Each rye B agar plate was inoculated with one disc of inoculum placed at the margin of the plate. Three tomato isolates and one potato isolate were used for characteristics. To determine the effect of temperature on the growth of each isolate, inoculated agar plates were incubated at 10, 12, 15, 18, 20, 24, 25, 26 and 27°C in darkness. The linear growth of mycelium was measured 10 days after inoculation. Four plates were used for each temperature, and the experiment was performed twice.

Determination of Mating Types

The mating type of each culture of P. infestans was determined by pairing a small piece (ca. 3 × 3 × 3 mm) of a culture to be tested with the same size of the A1 tester culture (isolate 533) or the A2 tester (isolate IB905) on a rye A agar block (ca. 15 × 10 × 3 mm) in a petri dish. Ten blocks were placed in a petri dish at equal distance along the edge. After incubation at 20°C in darkness for 10–14 days, agar blocks were examined microscopically. The isolates forming oospores when paired with the A2 tester were designated as A1. Similarly, isolates forming oospores when paired with A1 tester were designated A2.
Pathogenicity Tests

Inoculations were conducted using greenhouse-grown potato cultivar Kennebec and tomato cultivar Long-you 301 plants. Fully or nearly-fully expanded leaves were collected from the middle of the canopy. Non-terminal leaflets were placed in an inverted 150 × 15-mm petri dish mini moist chamber, which contained a layer of water agar on the top to provide moisture (Legard et al., 1995). Filter paper discs (Whatman No. 1, 9 mm in diameter) were saturated with about 0.3 ml of sporangial suspension of *P. infestans* (ca. 5 × 10⁴ sporangia /ml) and placed in the center of the lower leaf surface of each leaflet of potato and tomato plants, one disk per leaflet. Inoculated leaflets were moved into a moist room at 15–20°C in darkness with 95–100% relative humidity for disease development. Controls were inoculated with distilled water. Four replicate leaflets were used in each treatment. The experiments were conducted twice. Three tomato isolates, PIT-1, PIT-2, and PIT-3, isolated from Nantou, Taichung, and Chiayi, respectively, and two potato isolates, PIP-1 and PIP-2 isolated from Ching-jin Farm in Taichung County were used for pathogenicity tests.

Results

Mating Types and Geographical Distribution

All 68 isolates of *P. infestans* isolated from blighted tomato leaves collected from 12 different locations in the six counties of Taiwan were A1 (Table 1). Also, the mating types of the two isolates of *P. infestans* obtained from the blighted leaves of cultivated potato planted in the Ching-jin Farm in Taichung county were all A1 (Table 1). Neither A2 mating type nor self-fertile isolate was detected in Taiwan.

The geographical distribution of *P. infestans* isolates collected for this study is illustrated in Figure 1. Late blight on tomato usually occurs in high mountain areas (Nantou and Taichung counties) during wet summers. The disease has sometimes been observed in plain areas (Hualien, Ilan, Tainan and Chiayi counties) during the winter and spring. Generally, the disease is not common in Taiwan. Late blight was not observed in potato fields during this study.

![Figure 1. Geographic distribution of Phytophthora infestans in Taiwan.](image)

<table>
<thead>
<tr>
<th>Host</th>
<th>County</th>
<th>No. of fields</th>
<th>No. of isolates</th>
<th>Mating type</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>A1</td>
</tr>
<tr>
<td>Potato</td>
<td>Taichung</td>
<td>1</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Tomato</td>
<td>Ilan</td>
<td>3</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>Tomato</td>
<td>Hualien</td>
<td>1</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>Tomato</td>
<td>Taichung</td>
<td>3</td>
<td>24</td>
<td>24</td>
</tr>
<tr>
<td>Tomato</td>
<td>Nantou</td>
<td>2</td>
<td>21</td>
<td>21</td>
</tr>
<tr>
<td>Tomato</td>
<td>Chiayi</td>
<td>2</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>Tomato</td>
<td>Tainan</td>
<td>1</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>13</td>
<td>70</td>
<td>70</td>
</tr>
</tbody>
</table>
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Table 2. Characteristics of sporangia of Phytophthora infestans from tomato and potato.

<table>
<thead>
<tr>
<th>Host &amp; isolate</th>
<th>Location</th>
<th>Size of sporangia (µm)*</th>
<th>Length/width</th>
<th>Length of pedicel (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tomato</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PIT-1</td>
<td>Nantou</td>
<td>24.4–59.2 (38.2)</td>
<td>1.21–2.18 (1.56)</td>
<td>2.0–6.8 (3.56)</td>
</tr>
<tr>
<td>PIT-2</td>
<td>Taichung</td>
<td>25.2–48.0 (36.0)</td>
<td>1.31–2.40 (1.76)</td>
<td>0.8–4.8 (3.10)</td>
</tr>
<tr>
<td>PIT-3</td>
<td>Chiayi</td>
<td>19.2–52.0 (31.6)</td>
<td>1.10–2.60 (1.60)</td>
<td>1.2–4.4 (3.25)</td>
</tr>
<tr>
<td>Potato</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PIP-1</td>
<td>Taichung</td>
<td>18.0–40.0 (28.0)</td>
<td>1.22–2.0 (1.54)</td>
<td>2.0–4.0 (2.89)</td>
</tr>
</tbody>
</table>

*aMean dimensions are shown in brackets.

It was only found on the tissue of potted plants in the Ching-jin Farm in a high mountain area in Taichung County.

Characteristics of Phytophthora infestans

Sporangia of P. infestans were ovoid, ellipsoid to limoniform, tapering at the base, caducous, and semipapillate (Figure 2), ranging from 19.2–59.2 × 12–35.6 µm for isolates from tomato and 18–40 × 14–20.8 µm for the isolate from potato (Table 2). Sporangiophores were compound sympodial with a small swelling below the sporangium.

Two tested isolates of P. infestans, designated PIP-1 and PIT-1, obtained from potato and tomato, respectively, were able to grow on rye B agar plates from 10 to 25°C but not at and above 26°C (Figure 3). The optimum temperature for growth of both isolates was about 18–24°C.

Pathogenicity Tests

All three tomato isolates (PIT-1, PIT-2 and PIT-3) were able to cause leaf blight of tomato, but not of potato, while two potato isolates (PIP-1 and PIP-2) caused leaf blight on both potato and tomato. No disease symptoms were observed on control treatments. P. infestans was successfully re-isolated from all inoculated diseased plants.

Discussion

The presence of two mating types in heterothallic isolates of Phytophthora infestans is a prerequisite for their sexual reproduction and an indication of the origin of the species (Gallegly and Galindo, 1960; Fry et al., 1993; Ko, 1994). Before 1984, A2 mating type had only been detected in Mexico, the possible origin of P. infestans (Smoot et al., 1958). The A1 mating type prevailed in the rest of the world, including the United States, Canada, West Europe, South Africa, and West India (Smoot et al., 1958). Detection of the A2 mating type outside Mexico was first reported in 1984 from Switzerland (Hohl and Iselin, 1984), and subsequently from other places in Europe (Malcolmson, 1985; Tantius et al., 1986; Shaw et al., 1985), North America (Goodwin et al., 1994) and Asia (Mosa et al., 1989; So and Lee, 1993; Singh et al., 1994). Two hypotheses have been proposed to explain the new distribution of the A2 mating type. Fry et al. (1993) suggested that migration was the cause of the new occurrence of the A2 mating type, while Ko concluded in his research that sexual offspring or mutation from the descendants of A1 mating type pioneers could be the alternative origin (Ko, 1994). Recently, Goodwin and Drenth (1997) re-
analysed the published genotypic data to test the hypotheses on the origin of the A2 mating type outside Mexico. They concluded that the migration hypothesis was strongly supported and rejected Ko’s mating type change hypothesis. Long-distance migration of *P. infestans* frequently appears to have resulted from the inadvertent movement of infected plant material (potato tubers, tomatoes) during trade. The appearance of new populations of *P. infestans* has often been accompanied by devastating re-infestations. The appearance of new populations of infected plant material (potato tubers, tomatoes) during trade appears to have resulted from the inadvertent movement of *P. infestans*. Long-distance migration of *P. infestans* supported and rejected Ko’s mating type change hypothesis.

Tomato and potato are important vegetable crops in Taiwan, and both may be attacked by late blight. The isolates of *P. infestans* tested in the study all proved to be of the A1 mating type, as were thirteen isolates of *P. infestans* from Taiwan studied by Hartman and Huang (1995). This suggests that the Taiwanese *P. infestans* population may belong to the “old” *P. infestans* type, which was clonal and exclusively A1. However, the absence of the A2 mating type does not conclusively prove this, as some new populations lack A2 (Andrivon et al., 1994). To determine if the Taiwanese *P. infestans* population belongs to the old or new type, it will be necessary to characterise isolates using additional markers such as glucose-6-phosphate isomerase and aggressiveness (Andrivon et al., 1994; Goodwin et al., 1994; Lambert and Currier, 1997). If *P. infestans* in Taiwan indeed belongs to the old population type, it would be desirable to prohibit import of potato tubers from areas containing new populations of *P. infestans*, to avoid the possible destructive effect of the introduction of more aggressive forms of the pathogen.

The pathogenicity tests indicated a degree of host specialization in *P. infestans* in Taiwan. Isolates of *P. infestans* from tomato caused disease only in tomato, not potato, although the two potato isolates were able to infect both hosts. Host specialization to tomato has been reported in *P. infestans* from Taiwan studied by Hartman and Huang (1995). This suggests that the Taiwanese *P. infestans* population may belong to the “old” *P. infestans* type, which was clonal and exclusively A1. However, the absence of the A2 mating type does not conclusively prove this, as some new populations lack A2 (Andrivon et al., 1994). To determine if the Taiwanese *P. infestans* population belongs to the old or new type, it will be necessary to characterise isolates using additional markers such as glucose-6-phosphate isomerase and aggressiveness (Andrivon et al., 1994; Goodwin et al., 1994; Lambert and Currier, 1997). If *P. infestans* in Taiwan indeed belongs to the old population type, it would be desirable to prohibit import of potato tubers from areas containing new populations of *P. infestans*, to avoid the possible destructive effect of the introduction of more aggressive forms of the pathogen.

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**Literature Cited**


Phytophthora infestans


